Supplement of

Environmental DNA in different media reveals distribution characteristics and assembly mechanisms of fish assemblages in a complex river–lake system

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Table S1 The taxonomy and habitat water layer of fish detected in the Baiyang Lake via eDNA metabarcoding. The habitat water layer indicates which water layers the fish usually live. U: Upper layer, Lo: Lower layer, De: Demersal.

| Order	Family	Species	Traditional methods	eDNA	Habitat water layer
Cypriniformes	*Cyprinidae*	*Abbottina rivularis*	√	√	De
		Carassius auratus	√	√	Lo
		Ctenopharyngodon idella	√	√	Lo
		Culter alburnus	√	√	U
		Cyprinus carpio	√	√	Lo
		Chanodichthys erythropterus	√		U
		Hemiculter leucisculus	√	√	U
		Hypophthalmichthys molitrix	√	√	U
		Hypophthalmichthys nobilis	√	√	U
		Phoxinus oxycephalus jouyi		√	Lo
		Pseudorasbora parva	√	√	Lo
		Rhodeus ocellatus	√	√	Lo
		Zacco platypus		√	U
	Cobitidae	*Barbatula nuda*		√	De
		Lefua costata		√	De
		Cobitis macrostigma		√	De
		Misgurnus anguillicaudatus	√		De
		Misgurnus bipartitus		√	De
		Paramisgurnus dabryanus	√	√	De
	Acheilognathidae	*Acheilognathus macropterus*	√	√	U
	Leuciscidae	*Rhynchocypris lagowskii*		√	Lo
Perciformes	*Anabantidae*	*Macropodus chinensis*	√	√	Lo
	Channidae	*Channa argus*	√	√	De
	Gobiidae	*Ctenogobius giurinus*	√		De
		Rhinogobius brunneus		√	De
		Rhinogobius cliffordpopei		√	De
		Rhinogobius similis		√	De
	Mastacembelidae	*Mastacembelus armatus*		√	De
	Odontobutidae	*Micropercops swinhonis*	√	√	De
Silurfariformes	*Bngyidme*	*Pseudobagrus fulvidraco*	√	√	De
	Siluridae	*Silurus asotus*	√	√	De
Symbranchiformes	*Symbranchidae*	*Monopteras albus*	√		De
Beloniformes	*Adrianichthyidae*	*Oryzias latipes*		√	U

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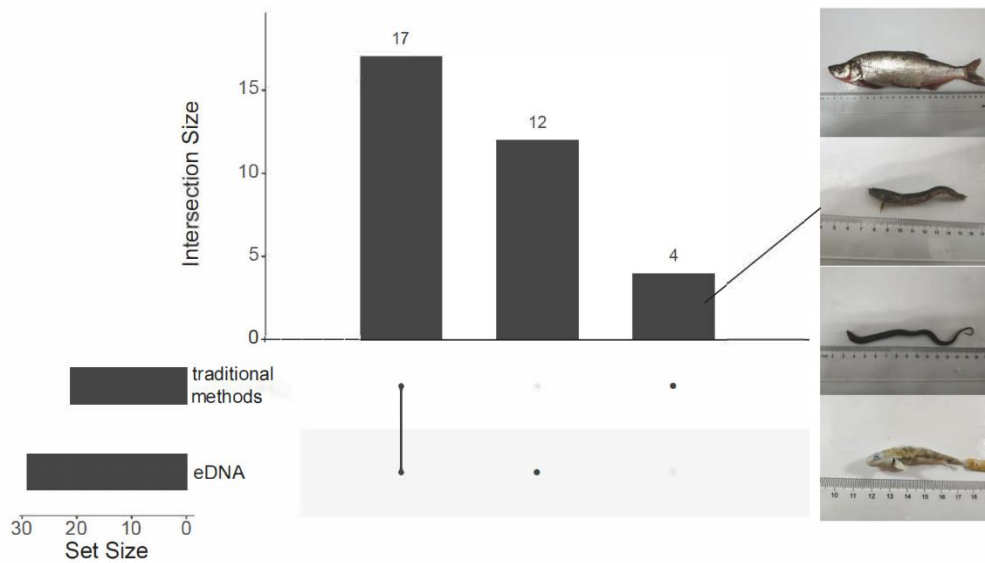
Figure S1 UpSet plots show the number of shared and different fish species detected by environmental DNA (eDNA) analyses and traditional surveys. The vertical bars with dots and lines below them show the fish species detected by both methods. The horizontal histograms next to the methods show the total number of fish species detected by each method. On the right are four species of fish detected only by morphology method, from top to bottom: *Chanodichthys erythropterus*, *Misgurnus anguillicaudatus*, *Ctenogobius giurinus*, and *Monopteras albus*.

Table S2 Names and distribution of species without recording in Baiyang Lake basin.

Species	Distribution
Amoya chusanensis	South China
Mastacembelus armatus	South China
Microphysogobio liaohensis	Nouth China
Odontobutis potamophila	South China
Channa punctata	South-East Asian
Lefua nikkonis	East Asian
Raneya brasiliensis	Marine
Trematomus bernacchii	Marine
Epinephelus akaara	Marine
Larimichthys crocea	Marine
Monodactylus argenteus	Marine
Zu cristatus	Marine
Odontobutis haifengensis	Marine
Epinephelus bruneus	Marine
Lutjanus campechanus	Marine
Takifugu rubripes	Marine
Regalecus glesne	Marine
Collichthys lucidus	Marine
Indostomus paradoxus	Marine
Myctophum affine	Marine
Scatophagus argus	Marine
Sparus aurata	Marine
Hippocampus queenslandicus	Marine

Pseudopentaceros richardsoni	Marine
Thamnaconus modestus	Marine
Banjos banjos	Marine
Tenualosa toli	Marine
Acipenser baerii	Marine
Aplocheilus lineatus	Marine
Neopomacentrus metallicus	Marine
Gudusia chapra	Marine
Ichthyborus	Marine

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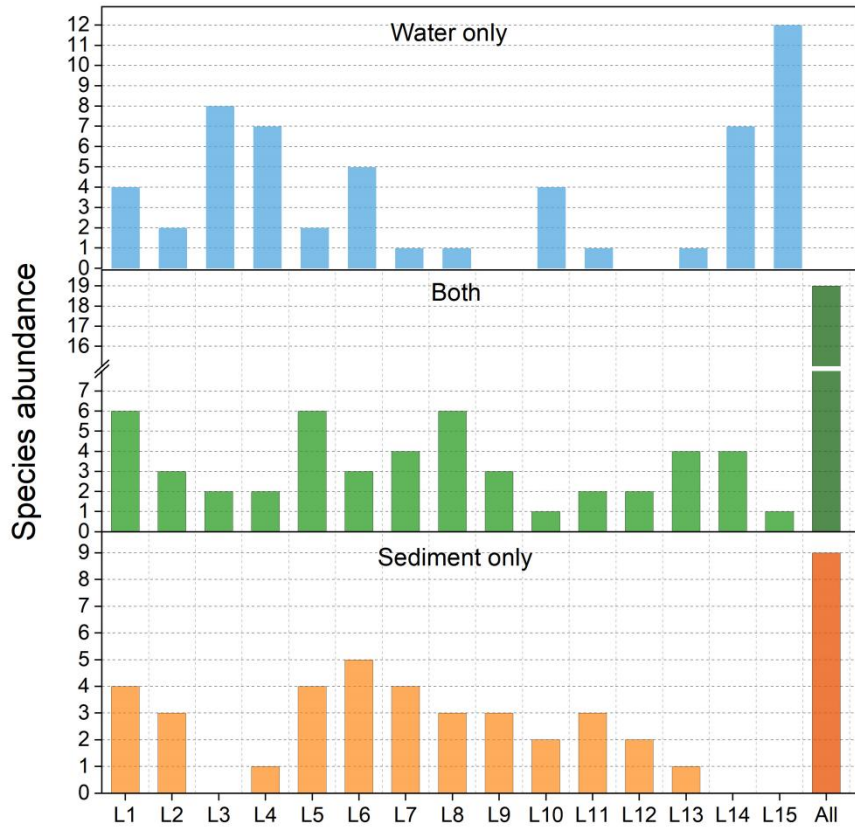
Figure S2 Comparison of the number of species detected in lake water and lake sediment samples. The species number detected in water only (upper histogram), both in water and sediment (middle histogram), and in sediment only (lower histogram) are shown. The last column is the sum of the 15 sites.

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Table S3 Variance inflation factor (VIF) and p-value of Monte Carlo permutation test for each environmental factor

Media type	Environmental factors	Variance inflation factor (VIF)	p-value
water	T	2.085731	0.025
	TP	1.892464	0.07
	DO	1.979852	0.085
	Chla	1.610442	0.085
	ORP	1.727767	0.25
	pH	2.26794	0.34
	SpCond	1.948048	0.4
	Turb	1.710016	0.775

	COD	2.759357	0.785
	TP	1.638360	0.015
	NH$_3$-N	2.379233	0.025
sediment	TN	1.659346	0.5
	Ni	2.277180	0.13
	Cr^{6+}	1.722893	0.745

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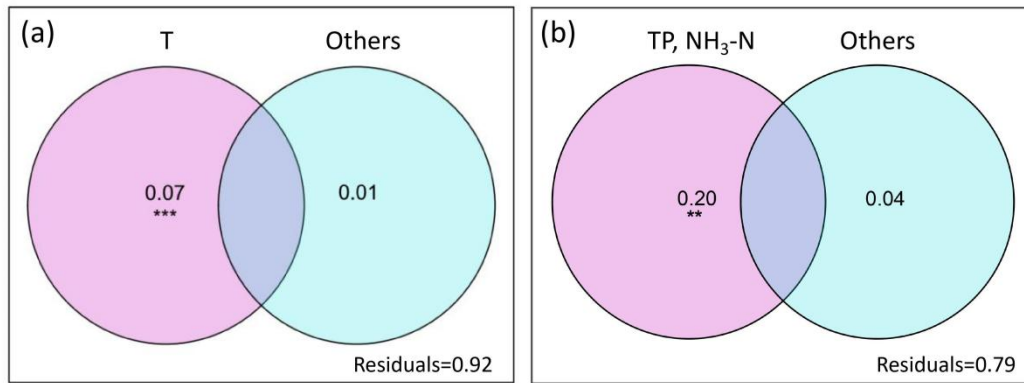
Figure S3 Variation partitioning analysis（VPA）of the contributions of environmental factors on the community structures in (a) water (river and lake) and (b) sediment (lake). The adjusted R^2 and dissimilarity significance are shown. **, $p < 0.01$; ***, $p < 0.001$, parts less than 0 are not displayed.

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